

## Article

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## Action of earthworms on flint burial – a return to Darwin's estate

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### Abstract

For thirty years, from the early 1840s, Charles Darwin documented the disappearance of flints in the grounds of Down House in Kent, at a location originally known as the “Stony Field”. This site (Great Pucklands Meadow - GPM) was visited in 2007 and an experiment set up in this ungrazed grassland. Locally-sourced flints (either large - 12 cm, or small – 5 cm dia.) were deposited at two densities within sixteen 1 m<sup>2</sup> plots in a randomised factorial design. The area selected was distant from public access routes and remained unmown throughout the duration here reported. Fixed point photographs were taken at the outset to enable later photogrammetric analysis. After 6 years, the site was re-examined. The flints had generally been incorporated into the soil. Photographs were re-taken, proportion of buried flints recorded and measurements made of burial depth from a quarter of each plot. Results showed that large flints were more deeply incorporated than smaller ( $p=0.025$ ), but more of the latter were below the soil surface. A controlled laboratory experiment was also conducted using *Aporrectodea longa* (the dominant earthworm species in GPM) to assess effects of casting in the absence of other biota. Results suggested that this species has a major influence on flint burial through surface casting. Combined with a long term, but small scale collection of *A. longa* casts from an area close to GPM, all results were consistent with those provided by Darwin and showed that rate of flint burial was within the range 0.21-0.96 cm y<sup>-1</sup>.

Keywords: bioturbation, Charles Darwin, casting, Down House, soil

## 1.0 Introduction

Charles Darwin lived at Down House for more than forty years and during that period undertook numerous experiments with earthworms that finally led to the production of “The Formation of vegetable mould through the action of worms” (FVM) (Darwin, 1881). This publication is now regarded by some as a cornerstone of the literature on pedogenesis, particularly so for first documenting the importance of faunal mixing of soil (Feller et al., 2003; Wilkinson et al., 2009; Johnson and Schaetzl, 2015). Darwin’s work on earthworms has been revisited numerous times over the last 130 years and has even formed the focus of conferences (e.g. Satchell, 1983). One relatively well known set of observations made by Darwin relate to a previously ploughed field (Great Pucklands Meadow (GPM)) that was left to become pasture when he purchased the property. His (1881) descriptions which relate to a period of 30 years are still as valuable today as when they were written:

“For several years it (GPM) was clothed with an extremely scant vegetation, and was so thickly covered with small and large flints (some of them half as large as a child’s head) that the field was always called by my sons “the stony field”. When they ran down the slope the stones clattered together, I remember doubting whether I should live to see these larger flints covered with vegetable mould and turf. But the smaller stones disappeared before many years had elapsed, as did every one of the larger ones after a time; so that after thirty years (1871) a horse could gallop over the compact turf from one end of the field to the other, and not strike a single stone with his shoes. To anyone who remembered the appearance of the field in 1842, the transformation was wonderful. This was certainly the work of the worms, for though castings were not frequent for several years, yet some were thrown up month after month, and these gradually increased in numbers as the pasture improved. In 1871 a trench was dug on the above slope...the turf was rather less than half an inch (1.27 cm), and the mould, which did not contain any stones, 2.5 inches (6.35 cm) in thickness. Beneath this lay coarse clayey earth full of flints, like that in any of the neighbouring ploughed fields. The average rate of accumulation of the mould during the whole thirty years was only .083 inch per year ( $0.21 \text{ cm y}^{-1}$ ) but the rate must have been much slower at first, and afterwards considerably quicker.”

This passage from FVM gives context to the work undertaken here. With permission from English Heritage, a field experiment was set up in Great Pucklands Meadow (GPM). The head Groundsman in 2007 gave assurances that every care would be taken to assist this work, which he and his successor kept to.

This work sought to add further data to information on the rate at which larger objects are buried by the action of earthworms – an action now termed bioturbation, which in a broad sense may be defined as the biological reworking of soils (and sediments) by all kinds of organisms (Meysman et al., 2006). From an earthworm perspective this may be considered to include burrowing through, ingesting and casting of soil. Original observations made by Darwin in GPM suggested that after cessation of ploughing, flints present on the soil surface were buried by natural processes over a period of years (above). Nevertheless, the assumption that this rate was not uniform cannot be substantiated due to the time intervals at which measurements were taken. A subsequent set of observations by Keith (1942), one hundred years after the first observations of Darwin's showed that the flints appeared to have come to rest on flinty-clay some 2.5 to 3 inches (6.35-7.62 cm) below an overlying stone-free earth.

The aim of this work was therefore to set up a long term field trial to replicate something of the situation which Darwin found in GPM when he moved to Down House in 1842 and record aspects of bioturbation. Objectives were to (a) record the rate of flint incorporation into the soil at this site; (b) compare incorporation of large and small flints, at high and low density; (c) determine the effects of flint burial by a selected earthworm species isolated from other soil macro-biota; (d) measure actual earthworm cast production in or close to GPM.

## **2 Materials and Methods**

The majority of the field work was undertaken within Great Pucklands Meadow (GPM) which is a part of the grounds of Down House, Kent, south east England. This was with the permission and collaboration of English Heritage, the owners and managers of the site. The soil type on site is described as freely draining, slightly acid loamy soil (NSRI, 2014). The pH is 5.6, OM content 5.2%,

and percentages of clay (21) silt (46) and sand (33). No stones greater than 2 mm were recorded in the surface soil (Beasley, Pers. Comm.). The field was completely covered with vegetation which had previously been sampled (NHM, 2006) to reveal a diverse sward with a species count of 119 flowering plants. An earthworm survey, using digging, hand-sorting of soil and vermifuge application (Butt et al., 2008) showed *Aporrectodea longa* (Ude) as the only casting species present in GPM, making up one fifth of the total density (290 m<sup>-2</sup>) and a half of the total biomass (115 g m<sup>-2</sup>). Three endogeic species were also present (*Allolobophora chlorotica* (Savigny), *Aporrectodea rosea* (Savigny), *Octolasion tyrtaeum* (Savigny)) and 2 epigeic species (*Lumbricus rubellus* (Hoffmeister) and *Satchellius mammalis* (Savigny)). *O. tyrtaeum* was the dominant earthworm species by number (59% of total density) (Butt et al., 2008).

## 2.1 Field Trial Establishment

In May 2007, flints were selected from a stockpile previously collected during routine works within the grounds of Down House. These were divided into 2 size classes – Large (L – approx. 12 cm dia. – mean mass 1,100 g) and Small (S – approx. 5 cm dia. – mean mass 250 g) these masses were derived from n= 5 of each, randomly selected for measurement. The flint size classes were a function of those available which naturally fell into two groups, lent themselves to inclusion in the experimental design and were reminiscent of the description given by Darwin (1881) of his own observations (see above). The location of the trial site (51°19'42.7"N 0°03'03.6"E) was determined to ensure that it would remain as undisturbed as possible for a lengthy duration. Although this work reports on findings after 6 years, it is planned to return on numerous occasions to continue similar monitoring for up to 20+ years. Care was taken to avoid a public footpath crossing GPM and also to avoid areas of known previous soil disturbance (e.g. Keith, 1942) where trenches had been dug and small areas of crops grown.

Within a square of 13 x 13 m, sixteen 1m<sup>2</sup> plots were separated by a distance of 3 m. A completely randomized 2 x 2 factorial arrangement was employed, to include the 2 flint size classes and 2 densities – High (HD) and Low (LD). For L and S flints respectively; HD = 50 or 100; LD = 25 or 50 m<sup>-2</sup>, with 4 replicates of each (see Table 1). Two flint densities were used as we hypothesised that the space occupied by flints could influence the subsequent filling of “non-flint” space. In other words, if there was less space for cast soil, the burial of the flints in the plot could be accelerated. The flints

were manually laid on the field surface directly on to existing relatively homogenous vegetation, dominated by grass species, but were not in contact with each other. After deposition, photographs were taken of each plot from 2 perspectives.

## 2.2 Field Trial Monitoring

After a period of 6 years, the site was re-visited, plots re-located and delineated with string using pegs previously left in place. The grass upon and around the plots was removed and dead vegetation around the flints was also carefully removed to allow a clear visual assessment of each plot. Any signs of surface earthworm casting were recorded along with any potential soil disturbance caused by other organisms. Photographs of each plot were taken as in 2007. In addition, some 20-30 images were taken encompassing a 360° view of each plot containing the larger size of flints, to permit photogrammetric analysis (e.g. Mikhail et al., 2001; RSPSOC, 2015). Photogrammetry permits measurements to be made from photographs, particularly to record the exact positions of surface points utilising numerous images taken from different angles of the same area.

Within each plot, the number of flints that could be seen at the soil surface was recorded. Each plot was then divided into quarters, with the same top right quarter, as viewed from Down House sampled for each plot. A visual assessment was made of each flint known to reside in that quarter (by reference to a 2007 photograph) with this non-normal data scored 1 if visible, and 0 if buried. Depth of flint burial within each quarter plot was then determined. Each flint was carefully removed from its position and the maximum depth below the soil surface measured manually against a probe held perpendicular to the soil surface. The flint was then marked, to ensure a single measurement for each, and replaced. If flints could not be seen, but were thought to be below the soil surface probing with a pointed needle was employed, and any found were extracted, depth measured and replaced as described above.

Vermifuge extraction (3 x 0.1 m<sup>2</sup>) was undertaken close to the experimental area to determine if the species of earthworm responsible for cast production, one aspect of bioturbation, was the same as that previously recorded (Butt et al., 2008). Five small hawthorn trees (*Crataegus monogyna*) that had begun to grow within the larger (13 x 13 m) square but not within the flint-filled plots, were dug up and removed in 2013.

### 2.3 Field assessment of cast production

Due to the length of unmown vegetation in GPM, it was thought to be difficult to collect quantitative data on earthworm cast production over lengthy periods. However, the earthworm species producing the majority of these casts at this location was *A. longa* and this species was also the dominant anecic in an adjacent regularly mown field (Great House Meadow – GHM; Butt et al., 2008).

Therefore to obtain an index of cast production at the soil surface, arrangements were made to have material collected by a visiting volunteer, from permanently defined 0.1 m<sup>2</sup> areas (n=10) every 2 weeks from April 2013 to March 2014. The castings were collected, bagged and sent to UCLan for dry mass determination. To verify that *A. longa* was responsible, vermifuge extraction below surface-casting was performed nearby, and confirmed previous sampling (Butt et al., 2008) where a community of earthworms similar to GPM was present, with *A. longa* as the dominant anecic.

### 2.4 Laboratory experiment

A controlled laboratory experiment was conducted using *A. longa* (the dominant earthworm species in GPM; Butt et al., 2008) to assess the effects of casting in the absence of other biota such as ants. Clear plastic, 2.5 litre pots with lids (Lakeland Plastics) had a standard soil added (Kettering loam, with pH 6.4, OM content 5%, and percentages of clay (24) silt (44) and sand (32)), and contents were then uniformly tapped to provide a standard pot depth (0.15 m), that may appear shallow for this species but has been used successfully in previous laboratory trials (Butt, 1993). A single small flint (obtained from the same stockpile as described above) was placed on the soil surface of each pot, ensuring that it did not touch the sides. Ten pots were kept as controls and a further 10 had 4 adult *A. longa* added. These animals were laboratory bred from stock sources using information derived from Butt (1993). All pots were initially provided with cut grass, but after 2 months with birch leaves. The change in food was decided upon as fungal growth was initially evident in the control pots. All pots were maintained in darkness at 15 °C (Lowe and Butt, 2005). At inspection, every 3 weeks, excess food material was removed, flints photographed from above and water spray provided, based on visual inspection of the soil surface. Fresh food was then re-supplied. The experiment was terminated after 24 weeks, at which point the depth of soil below the flints was determined by careful removal and probing with stiff wire to the base of the pot. In addition to this, the soil depth - mid-way along each side of the pot (n=4 pot<sup>-1</sup>) was measured from the outside. The survival and condition of

earthworms was also assessed. Comparisons of soil depths between experimental and control pots were then made.

## 2.5 Statistical analyses

With respect to depth of flint burial in the field, a standard 2 way analysis of variance was applied to the data gathered for stone size and density. For the binomial flint burial/non-burial data, it was appropriate to use an ANOVA with arcsine-square root transformed data. The laboratory experiment utilised student's t-tests to compare mean depth of soil below the flints and also mean soil depth at the pot margins. Field cast production was performed on a relatively small area (10 x 0.1 m<sup>2</sup>), so an overall assessment (total casting m<sup>-2</sup>) is presented.

## 3.0 Results

### 3.1 Field Trial

After 6 years, there was very little disturbance to the whole site and the majority of flints were still in positions as deposited. Only 3 large flints were located outside of the original plots at a maximum distance of 2.5 m, possibly moved by wild animals. All flints seemed to have been buried to some degree with the smaller completely buried in many cases. Figure 1a shows that large flints were found to be buried deeper than smaller. A 2-way ANOVA indicated significance of flint size (larger more deeply buried,  $p = 0.025$ ;  $F=6.505$ ;  $n= 4$ ), but not density, although there was a trend for high density flints to be buried more deeply (figure 1a). From the assessment of flints visible in the plots (Figure 1b), the data indicated that only a very few of the large flints were buried whereas up to 40% of small flints were belowground in 2013. Signs of earthworm casting were recorded in all of the sixteen plots and small scale indications of bioturbation by other soil fauna (red and black ant species) was recorded from 4 of the plots.

Multiple images were used to build a micro-topographic (photogrammetric point cloud) representation of the plots. In the example HDL plot from 2007, the warmer colours are "high" points, and cooler colours are lower. Individual yellow, orange and red mounds are evident for several of the flints (Figure 2a). The point cloud is not complete for these, as only 2 photographs were taken of each plot



in 2007 (e.g. Figure 2b), before considering photogrammetry would be a future possibility. After 6 years (2013), considerable smoothing of the surface (Figure 2c) was shown with only a minimal number of red peaks (from the same HDL plot No 9), notably, two of the flints towards the centre of the point cloud image, which correspond to the flints that are still clearly visible at the surface in Figure 2d. Note that in Figure 2c the point cloud is much denser than Figure 2a, due to 25+ photos being taken of each plot in 2013. Photogrammetry was only undertaken for large flint treatments (low and high density) in 2013, because the small flints were largely buried already.

### 3.2 Casting production

Figure 3 shows total cast production (dry mass) derived from a single year of data collected in Great House Meadow (GHM). A peak was observed in December, with little produced over the summer months of July and August. The overall production of cast material from the given square metre area equates to  $173 \text{ g m}^{-2} \text{ y}^{-1}$  (equivalent to  $1.73 \text{ t ha}^{-1} \text{ y}^{-1}$ ). Vermifuge application nearby revealed that the casting species was, as expected, *A. longa*. This was also the case in GPM, close to the experimental area.

### 3.3 Laboratory experiment

Casting was recorded within all of the pots containing earthworms and all earthworms survived until termination of the experiment. No casting or other bioturbation, such as burrowing occurred in the control pots. Mean measured soil depth at the margins of the pots was  $10.18 \pm 0.14 \text{ cm}$  in the presence of *A. longa* and  $9.48 \pm 0.12 \text{ cm}$  in the controls. This (0.7 cm) was significantly different ( $p=0.002$ ) but it is noted that edge effects are notorious in microcosm experiments with earthworms. By contrast, although differing by 0.39 cm, no significant difference ( $p=0.13$ ) was found between mean depth of the soil below the flints in the presence of *A. longa* ( $9.01 \pm 0.19 \text{ cm}$ ) or in the control pots ( $9.40 \pm 0.16 \text{ cm}$ ). Using these two separate measurements for effects of *A. longa* over 6 months, annual differences could be summed to give  $1.09 \text{ cm y}^{-1}$ . However, the density of *A. longa* in this experiment (pot area =  $12 \times 12 \text{ cm} = 144 \text{ cm}^2 = 1/70 \text{ m}^2$ ) is equivalent to  $(4 \times 70) 280 \text{ m}^{-2}$  which is 5.4 times greater than the field density recorded by Butt et al. (2008) for this species in GPM.

#### 4.0 Discussion

The almost complete lack of disturbance to the larger field plot showed that the site was well chosen and managed. The rate of incorporation over the first 6 years of the field trial was rapid, and greater than the average result ( $0.21 \text{ cm y}^{-1}$ ) supplied by Darwin (1881) and it was unexpected to find that many of the smaller flints had been fully buried. One major reason for this may have been that the current flints were laid on to living vegetation. It is likely that during the intervening six years that plant material died below the flints and formed a food source that was removed by the local earthworms and other detritivores and the greater mass of the larger flints may account for their recorded deeper incorporation. Also, Darwin had measured incorporation into what was initially a ploughed field with potentially fewer earthworms and lack of plant cover, so in 2007 by comparison, a fully developed earthworm community was already present. Further examination of flint depths after a similar time span, in for example 2019, will permit comparative rates of flint burial to be made under current field conditions. It will prove of some interest to see if the flints in our experiment are acted upon as those 130 years ago in this field and become static at the depth (2.5-3 inches) as recorded by Darwin and later by Keith (1942). It will also permit comparisons of incorporation rate since deposition.

Research by a number of authors, looking at incorporation of objects into the soil during e.g. ecological work (Yeates and Meulen, 1995) or e.g. archaeological investigations (Canti, 2003; Hanson et al., 2009) have shown that lumbricid earthworms have a major role in soil bioturbation and particularly soil surface burial of solid objects. Yeates and Meulen (1995) found that large wire rings had been incorporated at a rate of  $0.9\text{-}1.0 \text{ cm y}^{-1}$  in pasture where lumbricid density was in excess of  $1,000 \text{ m}^{-2}$ , with *Aporrectodea caliginosa* (Savigny), *L. rubellus* and *A. longa* present. Canti's (2003) review paper suggested that earthworms most definitely affect artefact burial and that, as might be predicted, the ecological group of earthworm species present on site and specific casting behaviour(s) are all relevant to burial rate. Nevertheless, the work also describes, as reported by Darwin (1881) and by Keith (1942) that "finds" and stones are often located at a particular horizon, despite the bioturbation brought about by earthworms. Hanson et al., (2009) developed the use of this process in a more applied setting and have shown, for example, that soil forensic investigations can utilise the action of earthworms to assist dating evidence burial.

The plots in GPM will be revisited after another 6 years have elapsed which will permit further photographic recording, to track progress of individual flints and even statistical comparisons of plot smoothness across temporal intervals, using photogrammetry. It is intended that monitoring will occur for at least a further three visits to utilise the unadulterated quarters of each plot, with returns thereafter still possible if flints can be re-interred without undue despoliation of the area.

Detail of the species of ants present will also be investigated, as particular species disturb soil more readily than others (e.g. Wilkinson et al., 2009). As no major anthills were recorded within any of the plots, it was not felt that this was a problem during current monitoring. However, the laboratory experiment showed that the effects of flint burial recorded from the field were in no doubt at least partially due to the effects of *A. longa* burrowing and casting. The experiment, whilst in a useful controlled setting, used small containers, held only one earthworm species and was only monitored for a relatively short period of six months. Calculations here, relating to actual flint burial, must therefore be considered with caution. The bioturbation observed will certainly be an overestimation of that from field conditions due e.g. to greater earthworm density, lack of rainfall, lower soil compaction and pot edge effects. Nevertheless, results did show significantly different soil levels brought about through earthworm casting compared with earthworm-free controls and were not too dissimilar to those provided by Darwin.

The casting results from Great House Meadow were valuable to show trends in production over an annual cycle, but as with any single year of study, these may be influenced by unseasonal occurrences, such as the cold spring and warm summer experienced in 2013, so this monitoring will also be continued for the foreseeable future. However, results obtained do mirror those of Evans and Guild (1947) as an index of earthworm activity across the year in Southern England. Differing management of GHM compared with GPM may not be significant, given similar numbers and biomasses of casting lumbricid species in both (Butt et al. 2008) but ideally data would have been collected directly from GPM.

The work described here could be viewed simply as monitoring flint burial over time, but can also be seen as a first step towards more fully documenting, at regular intervals, the long term observations made by Darwin in Great Pucklands Meadow. It is hoped that current values from laboratory and field,

recorded between 0.21 and 0.96 cm y<sup>-1</sup>, plus additional to be collected, may assist a more complete appreciation of bioturbation, brought about here by earthworms (and ants) in a historically significant setting that is well known to researchers from a host of scientific disciplines.

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## Figures

Figure 1 (a) Mean (+se) flint depth (cm) buried in the given treatments; (b) Proportion of flints totally buried in the given treatments based on count per plot.

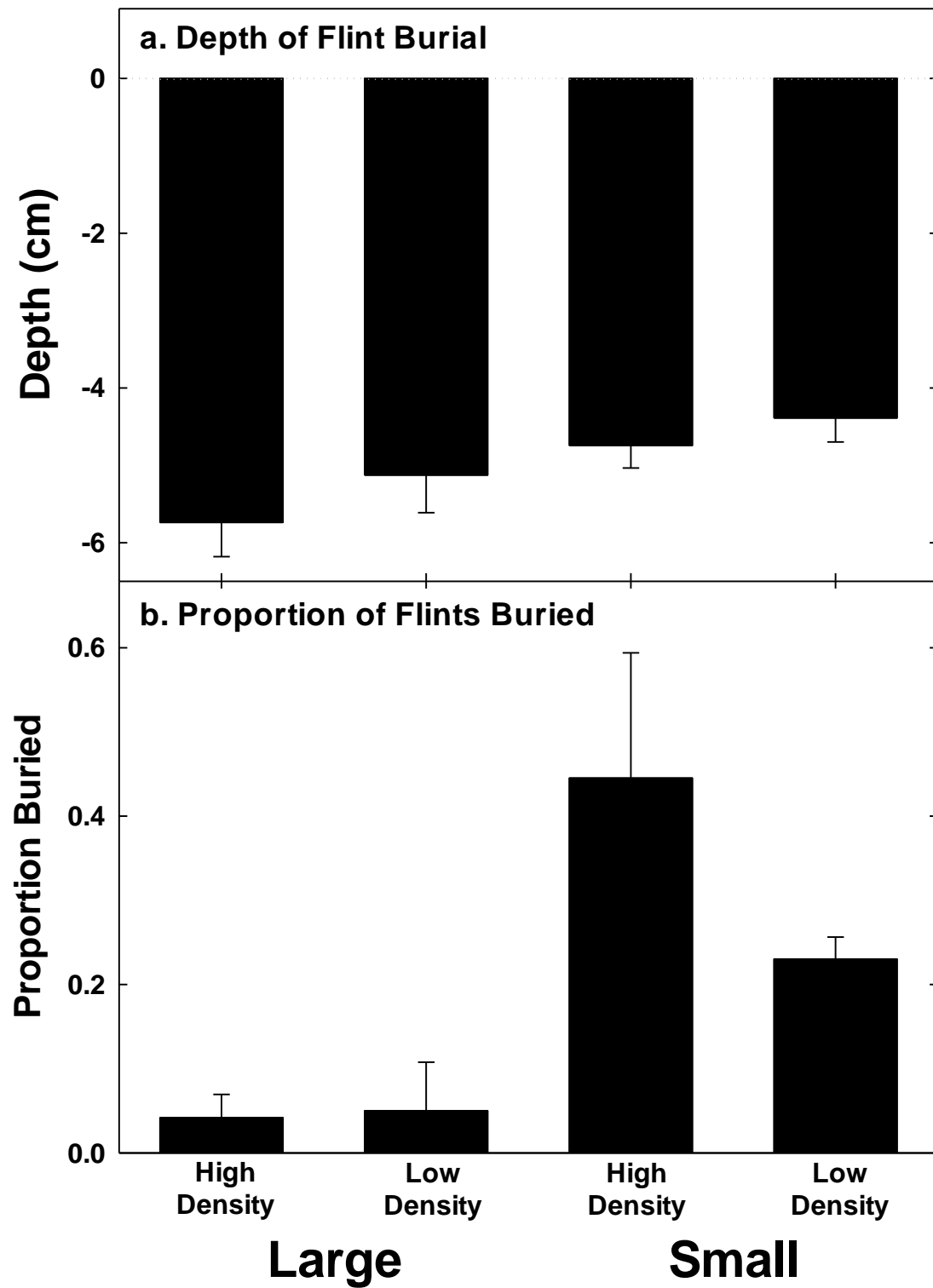
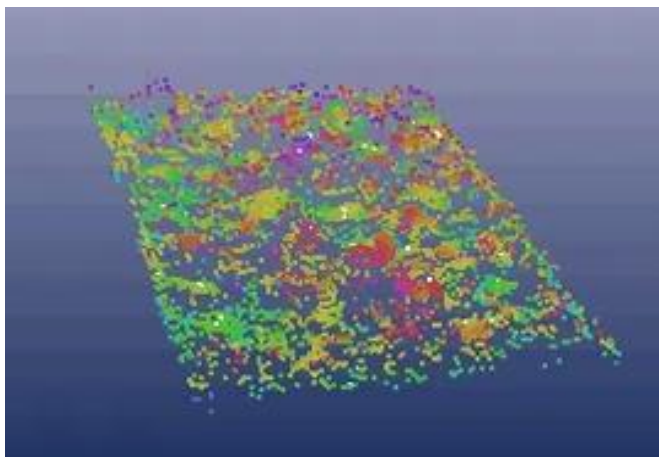


Figure 2 Photogrammetric (a) and (c) and photographic (b) and (d) representations of the same high density large (HDL) flint plot, in 2007 and 2013 respectively.

Figure 2

(a)

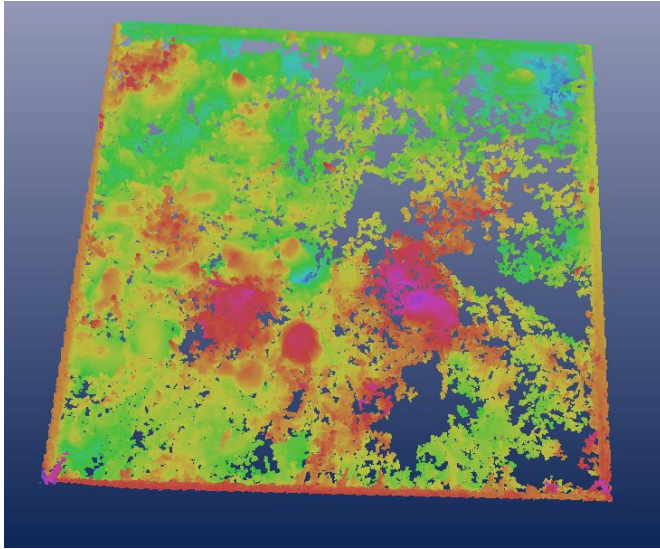


(b)





(c)



(d)



Figure 3 Annual earthworm cast production at Down House – derived from fortnightly collections in Great House Meadow (April 2013 – March 2014).

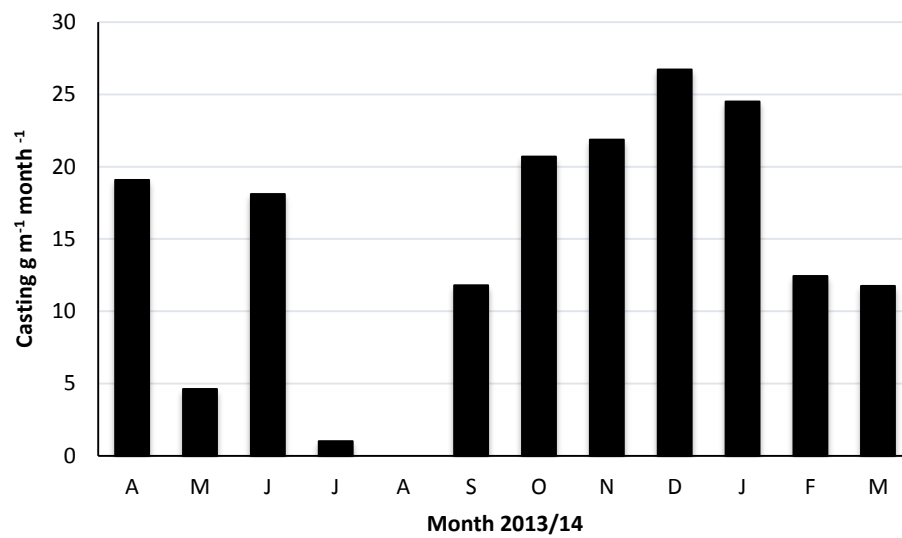


Table 1. Attributes of a field trial with 4 flint treatments in 1 m<sup>2</sup> plots (4 replicates) in Great Pucklands Meadow, Down House (HD – High Density; LD – Low Density; L – Large; S – Small).

Attribute	Treatment			
	HDL	LDL	HDS	LDS
No. Flints deposited plot <sup>-1</sup> (2007)	50	25	100	50
Mean ± se flint Mass (g)	1,100 ± 160	1,100 ± 160	250 ± 37	250 ± 37
Mean ± se No. flints visible at surface (2013)	36.75 ± 3.59	22.5 ± 1.89	24 ± 6.36	22 ± 3.34
(% of original)	(74)	(90)	(24)	(44)
Mean ± se burial depth (cm)	5.74 ± 0.18	5.12 ± 0.23	4.74 ± 0.12	4.39 ± 0.17
Burial rate (cm y <sup>-1</sup> )	0.96	0.85	0.79	0.73